



**Whole Effluent Toxicity Test Report:
Washington Beef LLC.**

June 2013

Report date: July 3, 2013

Submitted to:

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1.0 INTRODUCTION

A whole effluent toxicity test was conducted using effluent samples collected from the Washington Beef LLC wastewater treatment plant in June 2013. A chronic bioassay was conducted using the test organism *Ceriodaphnia dubia* (*Ceriodaphnia*). Testing was performed at Rainier Environmental Laboratory located in Tacoma, Washington.

2.0 METHODS

2.1 Sample Collection and Transport

Effluent samples were collected into 4-liter (L) LDPE cubitainers by Washington Beef personnel. The samples were packed in coolers containing ice and shipped to Rainier Environmental by overnight delivery service. Appropriate chain-of-custody procedures were employed during collection and transport (Appendix D).

2.2 Sample Receipt

Upon arrival at the laboratory, coolers were opened, samples inspected, and the contents verified against information provided on the chain-of-custody forms. Receipt temperature was measured and recorded on the chain-of-custody form. The standard water quality parameters were measured and recorded on sample check-in sheets (Appendix B). Samples were stored at 4°C in the dark until used for testing.

2.3 Test Methods

A chronic toxicity test was conducted according to procedures presented by USEPA (2002). The methods are summarized in Table 1. The procedure involved a 7-day static-renewal exposure to the effluent. The endpoints from these tests were *Ceriodaphnia* survival at the end of exposure and reproduction at test termination or production of 3 broods, whichever occurred first. Termination of the test occurred when at least 60 percent of surviving control females produced 3 broods. The test was ended on Day 7.

Table 1. Summary of methods for the 7-day *Ceriodaphnia* survival and reproduction test.

Test initiation date and time	6/18/13; 1530h
Test termination date and time	6/25/13; 1540h
Test organism	<i>Ceriodaphnia dubia</i>
Test organism source	In-house cultures
Test organism age	< 24 hours
Test duration	7 days; Test terminated when 60% of controls reached 3 broods
Feeding	1:1 mixture YTC:algal suspension daily
Test chamber; test solution volume	30 mL plastic cup; 15 mL
Test temperature	25 ± 1°C
Dilution water	Diluted mineral water
Test concentrations (% sample)	100, 50, 25, 12.5, 6.25, laboratory control
Number of organisms/chamber	1
Number of replicates	10
Photoperiod	16 hours light/8 hours dark
Aeration	None
Test protocol	EPA-821-R-02-013
Test acceptability criteria for controls	≥ 80% survival; ≥ 15 neonates per surviving adult
Reference toxicant	Sodium chloride

3.0 RESULTS AND DISCUSSION

Details of standard water quality measurements conducted upon receipt of samples are provided in Table 2.

Table 2. Final Effluent sample information.

Parameter	F.E Outfall 002		
Rainier Log-in No.	13-061	13-064	13-066
Collection date and time	6/17/2013; 0715h	6/19/2013; 0715h	6/21/2013; 0720h
Receipt date and time	6/18/2013; 1415h	6/20/2013; 1400h	6/22/2013; 1040h
Receipt temperature (°C)	1.0	1.0	1.5
Dissolved oxygen (mg/L)	9.0	7.8	7.5
pH	7.70	7.82	7.73
Conductivity (µS/cm)	4820	4160	4550
Salinity (ppt)	2.4	2.2	2.4
Hardness (mg/L CaCO ₃)	60	68	72
Alkalinity (mg/L CaCO ₃)	120	144	144
Total Chlorine (mg/L) ^a	<0.03	<0.03	<0.03
Total Ammonia (mg/L) ^b	<1.0	<1.0	<1.0

^{a,b} See reference below

Note: Total chlorine and ammonia values are measured by Rainier Environmental to provide additional information in support of the bioassay test procedures. They are not intended to be interpreted as exact values, particularly near the detection limits where interferences are most likely to become apparent.

^a Total chlorine is measured using a Hach DR/2000 spectrophotometer and colorimetric DPD Total Chlorine Reagent. Under optimum conditions, the method has a range of 0.03 to 2.0 mg/L \pm 0.01 mg/L total chlorine. Compounds in the sample that interfere with chlorine detection include bromine, manganese, chromium, ozone, and peroxides. Additional interferences include extreme pH values and high alkalinity (greater than 300 mg/L Ca CO₃).

^b Total ammonia is measured using a Hach DR/2000 spectrophotometer following the salicylate method which uses AmVer Diluent Reagent Test 'N' Tube kits. Under optimum conditions, the method has a range of 0.4 to 50.0 \pm 0.1 mg/L NH₃-N. High sample turbidity will give erroneously high values. Additional interferences to the method include extreme pH and high concentrations of magnesium, iron, nitrite, nitrate, or sulfate.

Results for the toxicity tests are summarized in Table 3. Individual statistical summaries for the test and copies of the laboratory bench sheets are provided in the Appendices A-D.

The NOEC (concentration at which no effect on the organisms is detected) was 50 percent sample for survival and 25 percent for reproduction. The associated chronic toxicity unit (TUC; 100 percent sample divided by the NOEC) was 2 for survival and 4 for reproduction.

Table 3. Summary of toxicity test results.

Sample	Endpoint	NOEC (% effluent)	Chronic Toxicity Unit (TUC) ^a
Final Effluent	Survival	50	2.0
	7-day Reproduction	25	4.0

^a Chronic toxicity unit (TUC = 100 \div NOEC)

4.0 QA/QC

Samples were received in good condition and within the temperature range specified by EPA (2002). The toxicity tests met all acceptability criteria for performance of control organisms. There were no deviations from protocol and water quality parameters remained within the ranges specified in the corresponding test methods throughout the tests.

Results for the most recent reference toxicant test used to monitor laboratory performance and test organism sensitivity are summarized in Table 4 and Appendix C. The coefficients of variation (CV) for the endpoints are also shown in the table. The results for the reference toxicant test fell within the acceptable range of mean \pm two standard deviations of historical test results indicating that the test organisms were of an appropriate degree of sensitivity.

Table 4. Reference toxicant test results.

Species	Endpoint	Date initiated	LC ₅₀ /EC ₅₀	Acceptable Range	CV (%)
<i>Ceriodaphnia</i>	7d survival	6/18/2013	1.87 g/L NaCl	1.29 – 2.88 g/L	22.2
	7d reproduction	6/18/2013	1.26 g/L NaCl	1.07 - 1.60 g/L	10.5

REFERENCES

Tidepool Scientific Software. 2000-2010. CETIS Comprehensive Environmental Toxicity Information System Software, Version 1.8.0.8.

USEPA. 2002. Short-term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Freshwater Organisms, Fourth Edition. EPA-821-R-02-013. pp. 141-196.

Appendix A
Statistical Summaries and Raw Bench Sheets

CETIS Summary Report

Report Date: 03 Jul-13 14:54 (p 1 of 2)
 Test Code: 1306-027 | 03-5484-6978

Ceriodaphnia 7-d Survival and Reproduction Test				Rainier Environmental Laboratory		
Batch ID:	02-7452-6100	Test Type: Reproduction-Survival (7d)				Analyst: Eric Tollefson
Start Date:	18 Jun-13 15:30	Protocol: EPA/821/R-02-013 (2002)				Diluent: Perrier Water
Ending Date:	25 Jun-13 15:40	Species: Ceriodaphnia dubia				Brine:
Duration:	7d 0h	Source: In-House Culture				Age: <24h
Sample ID:	19-6736-8054	Code:	13-061	Client: Washington Beef		
Sample Date:	17 Jun-13 07:15	Material:	POTW Effluent	Project:		
Receive Date:	18 Jun-13 14:15	Source:	Washington Beef (WA0050202)			
Sample Age:	32h (1 °C)	Station:	Outfall 002			

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
06-3793-3487	7d Survival Rate	50	100	70.71	NA	2	Fisher Exact/Bonferroni-Holm Test
03-2365-6760	Reproduction	25	50	35.36	17.0%	4	Steel Many-One Rank Sum Test

Point Estimate Summary

Analysis ID	Endpoint	Level	%	95% LCL	95% UCL	TU	Method
08-8531-9748	7d Survival Rate	LC5	52.55	51.77	54.55	1.903	Linear Interpolation (ICPIN)
		LC10	55.23	53.61	59.5	1.811	
		LC15	58.04	55.5	64.89	1.723	
		LC20	60.99	57.47	70.77	1.639	
		LC25	64.1	59.5	77.17	1.56	
		LC40	74.36	66.03	100	1.345	
03-8585-9370	Reproduction	LC50	82.09	70.77	N/A	1.218	Linear Interpolation (ICPIN)
		IC5	18.47	1.537	26.44	5.415	
		IC10	25.38	5.438	28.23	3.941	
		IC15	27.26	16.12	30.1	3.669	
		IC20	29.27	22.14	32.22	3.416	
		IC25	31.43	25.91	34.52	3.182	
		IC40	38.87	34.1	43.25	2.572	
		IC50	44.76	39.1	50.5	2.234	

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits	Overlap	Decision
06-3793-3487	7d Survival Rate	Control Resp	1	0.8 - NL	Yes	Passes Acceptability Criteria
08-8531-9748	7d Survival Rate	Control Resp	1	0.8 - NL	Yes	Passes Acceptability Criteria
03-2365-6760	Reproduction	Control Resp	35.2	15 - NL	Yes	Passes Acceptability Criteria
03-8585-9370	Reproduction	Control Resp	35.2	15 - NL	Yes	Passes Acceptability Criteria
03-2365-6760	Reproduction	PMSD	0.1696	0.13 - 0.47	Yes	Passes Acceptability Criteria

7d Survival Rate Summary

C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Dilution Water	10	1	1	1	1	1	0	0	0.0%	0.0%
		10	1	1	1	1	1	0	0	0.0%	0.0%
		10	1	1	1	1	1	0	0	0.0%	0.0%
		10	1	1	1	1	1	0	0	0.0%	0.0%
		10	1	1	1	1	1	0	0	0.0%	0.0%
		10	0.3	0.1196	0.4804	0	1	0.1528	0.483	161.0%	70.0%

Reproduction Summary

C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Dilution Water	10	35.2	32.64	37.76	17	40	2.164	6.844	19.44%	0.0%
		10	35.5	32.93	38.07	18	43	2.177	6.884	19.39%	-0.85%
		10	35.4	33.06	37.74	19	40	1.979	6.257	17.68%	-0.57%
		10	32.2	30.06	34.34	17	39	1.812	5.731	17.8%	8.52%
		10	14.9	12.66	17.14	2	22	1.9	6.008	40.32%	57.67%
		10	0.4	-0.07233	0.8723	0	4	0.4	1.265	316.2%	98.86%

CETIS Summary Report

Report Date:

03 Jul-13 14:54 (p 2 of 2)

Test Code:

1306-027 | 03-5484-6978

Ceriodaphnia 7-d Survival and Reproduction Test

Rainier Environmental Laboratory

7d Survival Rate Detail

C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Dilution Water	1	1	1	1	1	1	1	1	1	1
6.25		1	1	1	1	1	1	1	1	1	1
12.5		1	1	1	1	1	1	1	1	1	1
25		1	1	1	1	1	1	1	1	1	1
50		1	1	1	1	1	1	1	1	1	1
100		0	1	0	0	0	1	0	0	0	1

Reproduction Detail

C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Dilution Water	40	39	40	38	32	38	37	36	35	17
6.25		38	39	39	39	32	38	43	35	34	18
12.5		39	38	40	38	32	40	37	36	35	19
25		32	39	33	35	35	33	32	33	33	17
50		17	22	16	19	14	2	17	12	21	9
100		0	0	0	0	0	0	4	0	0	0

7d Survival Rate Binomials

C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Dilution Water	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
6.25		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
12.5		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
25		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
50		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
100		0/1	1/1	0/1	0/1	0/1	0/1	1/1	0/1	0/1	1/1

Rainier Environmental
Washington Laboratory

Client: Washington Beef
Sample ID: F.E. OUTFALL CO2
Test No: 1306-027
Log-In#: 13-061 13-064

Initial and Final Chemistries

Seven Day Chronic Freshwater Bioassay

Start Date & Time: 6/18/13 1530

Stop Date & Time: 6/25/13 1540

Test Species: Ceriodaphnia dubia

13-066

Conc. or % CON	Days													
	0		1		2		3		4		5			
	init.	final	init.	final										
pH	7.60	7.80	7.43	7.84	7.62	7.91	7.61	7.93	7.75	8.01	7.82	8.01	7.69	8.00
DO (mg/l)	7.3	7.8	7.7	7.9	7.5	8.0	8.1	8.2	8.1	8.2	8.1	8.1	8.0	8.2
Cond. (umhos-cm)	300	190	195	205	203	207	196	200	203	208	199	212	201	215
Temperature (°C)	26.0	24.7	25.3	25.1	24.7	25.2	25.1	25.1	25.1	25.4	24.7	25.5	24.9	25.4
6.25	Days													
	0		1		2		3		4		5			
	init.	final	init.	final										
pH	7.63	7.81	7.52	7.93	7.67	7.94	7.65	7.97	7.72	8.03	7.80	7.95	7.63	7.97
DO (mg/l)	7.3	7.8	7.9	7.9	7.7	7.8	8.0	8.1	8.1	8.2	8.1	8.1	8.0	8.1
Cond. (umhos-cm)	503	510	506	528	506	523	502	511	501	522	505	528	507	519
Temperature (°C)	25.0	24.6	25.3	26.2	24.8	25.5	25.1	25.1	25.0	25.4	24.9	25.5	24.7	25.3
12.5	Days													
	0		1		2		3		4		5			
	init.	final	init.	final										
pH	7.64	7.83	7.54	7.94	7.69	7.97	7.65	8.03	7.67	8.05	7.79	7.93	7.63	7.98
DO (mg/l)	7.6	7.9	8.0	7.9	7.9	7.8	7.7	8.1	8.1	8.2	8.2	8.1	8.2	8.2
Cond. (umhos-cm)	903	922	917	954	913	927	904	919	918	916	905	923	899	913
Temperature (°C)	25.1	24.6	25.3	25.2	25.1	25.4	25.0	25.1	25.0	25.4	24.8	25.5	24.7	25.3
25	Days													
	0		1		2		3		4		5			
	init.	final	init.	final										
pH	7.69	7.89	7.57	8.01	7.68	7.89	7.69	8.05	7.63	8.11	7.62	7.97	7.60	8.00
DO (mg/l)	7.8	7.6	8.0	8.2	8.0	7.9	7.7	8.3	8.0	8.2	8.2	8.3	8.1	8.2
Cond. (umhos-cm)	1492	1515	1497	1575	1507	1551	1481	1532	1475	1503	1491495	1515	1505	1528
Temperature (°C)	25.3	24.6	25.2	25.2	25.4	25.4	24.8	25.1	25.3	25.4	25.0	25.5	25.0	25.4
50	Days													
	0		1		2		3		4		5			
	init.	final	init.	final										
pH	7.60	7.97	7.55	8.04	7.67	8.03	7.61	8.12	7.59	8.13	7.59	8.01	7.56	8.11
DO (mg/l)	7.9	8.2	8.0	7.8	8.1	7.8	8.0	8.2	8.0	8.2	8.2	8.1	8.2	8.1
Cond. (umhos-cm)	2638	2636	2652	2747	2684	2723	2617	2671	2498	2558	2598	2617	2607	2638
Temperature (°C)	25.4	24.8	25.1	25.3	25.2	25.2	24.5	25.1	25.4	25.4	25.1	25.5	25.1	25.4
100	Days													
	0		1		2		3		4		5			
	init.	final	init.	final										
pH	7.53	8.04	7.47	8.17	7.57	8.11	7.54	8.17	7.46	8.21	7.52	8.10	7.52	8.13
DO (mg/l)	8.2	7.4	8.1	7.8	8.2	7.5	8.2	8.2	7.8	8.2	8.1	8.2	8.2	8.1
Cond. (umhos-cm)	4940	4910	4990	5040	4960	5010	4910	4930	4790	4950	4910	5030	4970	4930
Temperature (°C)	25.8	24.9	25.0	25.3	25.2	25.1	24.3	25.1	25.7	25.4	25.4	25.5	25.4	25.4
Tech. Initials	BT	BT	BT	BT										

Dilution Water Batch #: BMW 004

Test Chamber: VWR

QA Check: BT

Sample Description:

Animal Source: In House Culture

Date Received:

Date of Hatch:

Comments:

Ceriodaphnia 7-Day Chronic Survival and Reproduction

Start Date and Time:
6/18/13 15:30

Stop Date and Time:
6/25/13 15:40

Client/Sample ID: WA_BEEF
Test Number: 1306-027

Rep	Conc.	Cont	Daily Reproduction								Day 6 Total	Third Brood	
			1	2	3	4	5	6	7	8			
1	CON	19	=	=	=	5	=	14	31	19	40	1	25
2		27	=	=	=	6	=	13	20	19	39	2	11
3		29	=	=	=	4	=	14	22	18	40	3	53
4		57	=	=	=	5	=	13	20	18	38	4	33
5		3	=	=	=	5	=	12	15	17	32	5	41
6		48	=	=	=	5	=	13	21	17	39	6	13
7		49	=	=	=	6	=	13	18	19	37	7	23
8		54	=	=	=	5	=	11	30	16	36	8	45
9		15	=	=	=	6	=	14	15	20	35	9	43
10		10	=	=	=	5	=	13	—	17	17	10	34
Analyst			X	Q	Q	Q	Q	Q	Q	Q	Q		
Time			15:30	10:30	9:00	10:40	10:45	10:50	11:00	13:40			
Selen #			003	003	003	003	003	003	003	003			
Rep	Conc.	Cont	Day 6 Third Brood								Day 6 Total	Third Brood	
			1	2	3	4	5	6	7	8			
1	6.35	30	=	=	=	5	=	12	31	17	39	1	50
2		44	=	=	=	6	=	13	20	19	39	2	26
3		37	=	=	=	6	=	13	20	19	39	3	26
4		16	=	=	=	6	=	13	20	19	39	4	56
5		1	=	=	=	6	=	11	15	17	32	5	4
6		7	=	=	=	5	=	12	21	17	38	6	39
7		50	=	=	=	5	=	16	22	21	43	7	17
8		46	=	=	=	5	=	13	17	18	35	8	30
9		59	=	=	=	5	=	13	16	18	34	9	21
10		3	=	=	=	5	=	13	13	18	16	10	32

Rep	Conc.	Cont	Day 6 Third Brood								Day 6 Total	Third Brood	
			1	2	3	4	5	6	7	8			
1	13.5	36	=	=	=	5	=	13	21	18	37	1	100
2		32	=	=	=	6	=	12	20	18	38	2	31
3		53	=	=	=	6	=	14	20	20	40	3	35
4		58	=	=	=	5	=	13	20	19	38	4	25
5		58	=	=	=	5	=	12	15	17	33	5	47
6		12	=	=	=	6	=	15	19	21	40	6	6
7		14	=	=	=	6	=	11	13	19	37	7	9
8		18	=	=	=	5	=	13	17	19	36	8	24
9		2	=	=	=	5	=	13	17	18	35	9	40
10		43	=	=	=	6	=	13	13	15	39	10	60

Rep	Conc.	Cont	Daily Reproduction								Day 6 Total	Third Brood	
			1	2	3	4	5	6	7	8			
1	25	51	=	=	=	5	=	11	16	16	32	1	25
2		11	=	=	=	6	=	13	20	19	39	2	53
3		53	=	=	=	5	=	12	16	17	33	3	33
4		33	=	=	=	5	=	12	18	19	35	4	41
5		13	=	=	=	7	=	12	16	16	33	5	13
6		7	=	=	=	7	=	11	11	15	33	6	23
7		7	=	=	=	7	=	11	11	15	33	7	17
8		7	=	=	=	6	=	14	7	10	17	8	17
9		7	=	=	=	6	=	14	7	10	17	9	12
10		7	=	=	=	5	=	13	11	14	21	10	17

Comments: X=mortality

Appendix B
Sample Check-In Sheets

Rainier Environmental
5013 Pacific Hwy East, Ste. 20
Tacoma, WA 98424

Sample Check-In Information

Client: Washington Bell

Test ID No(s.): Cd-C
1306-027

Sample Description:
Brown

Sample ID:	Final Effluent	F.E. outlet 100A	F.E. outlet 100B	
Login No. (10-xxxx):	13-061	13-064	13-066	
Sample Collection Date & Time:	6/17/13 7:15	6/19/13 7:15	6/6/13 7:20	
Sample Receipt Date & Time:	6/18/13 14:15	6/20/13 14:00	6/21/13 10:40	
Check-in Temperature (°C)	1.0	1.0	1.5	
Temperature OK?	Y N	Y N	Y N	
DO (mg/L)	9.0	7.8	7.5	
pH (units)	7.70	7.82	7.73	
Conductivity (µS/cm)	4820	460	4550	
Salinity (ppt)	2.4	2.2	2.4	
Tit. Vol / Sam. Vol. / Alkalinity (mg/gL)*	3.0 / 2.5 / 120	3.6 / 2.5 / 144	3.6 / 2.5 / 144	/ /
Tit. Vol. / Sam. Vol. / Hardness (mg/gL)* ^a	1.5 / 2.5 / 60	1.7 / 2.5 / 63	1.8 / 2.5 / 72	/ /
Total Chlorine (mg/L)	<0.03	<0.03	<0.03	
Total Ammonia (mg/L)	<1.0	<1.0	<1.0	
Technician Initials	ET	94	94	

* = mg/L as CaCO₃, ^a = Measured for freshwater samples only, NA = Not Applicable,

NM = Not Measured

Freshwater Tests:

Control/Dilution Water Source: test type: cd-c 8.2 (DMW) MHW Other: _____ Alkalinity: 64 Hardness: 86

Control/Dilution Water Source: test type: _____ 8.2 (DMW) MHW Other: _____ Alkalinity: _____ Hardness: _____

Additional Control? Y N = _____ Alkalinity: _____ Hardness: _____

Marine Tests:

Control/Dilution Water Source: test type: _____ ART SW NAT SW Alkalinity: _____ Salinity: _____

Control/Dilution Water Source: test type: _____ ART SW NAT SW Alkalinity: _____ Salinity: _____

Additional Control? Y N = _____ Alkalinity: _____ Salinity: _____

Sample Salted w/ artificial salt? Y N If yes, what ppt? _____ test type: _____

Sample salted w/brine? Y N If yes, what ppt? _____ test type: _____

Comments: Temperature for grab sample must be 0-20°C if received within 1 hour of collection time, 0-12°C if effluent received within 4 hours of collection time, and 0-6°C for all other samples.

QC Check: 94

Hardness Adjustment? Y N
If adjusted, please see worksheet for details.

Sub-samples for additional chemistry:

Appendix C
Reference Toxicant Test

Ceriodaphnia 7-d Survival and Reproduction Test

Rainier Environmental Laboratory

Test Type: Reproduction-Survival (7d)

Organism: Ceriodaphnia dubia (Water Flea)

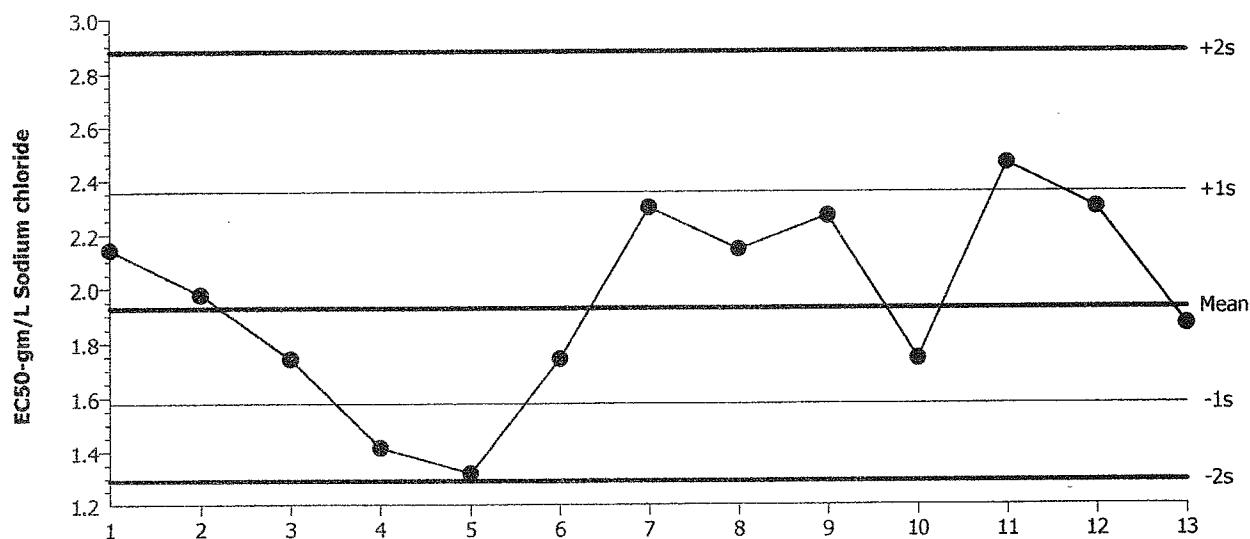
Material: Sodium chloride

Protocol: EPA/821/R-02-013 (2002)

Endpoint: 7d Survival Rate

Source: Reference Toxicant-REF

Ceriodaphnia 7-d Survival and Reproduction Test



Mean: 1.928

Count: 12

-1s Warning Limit: 1.578

-2s Action Limit: 1.291

Sigma: NA

CV: 22.20%

+1s Warning Limit: 2.356

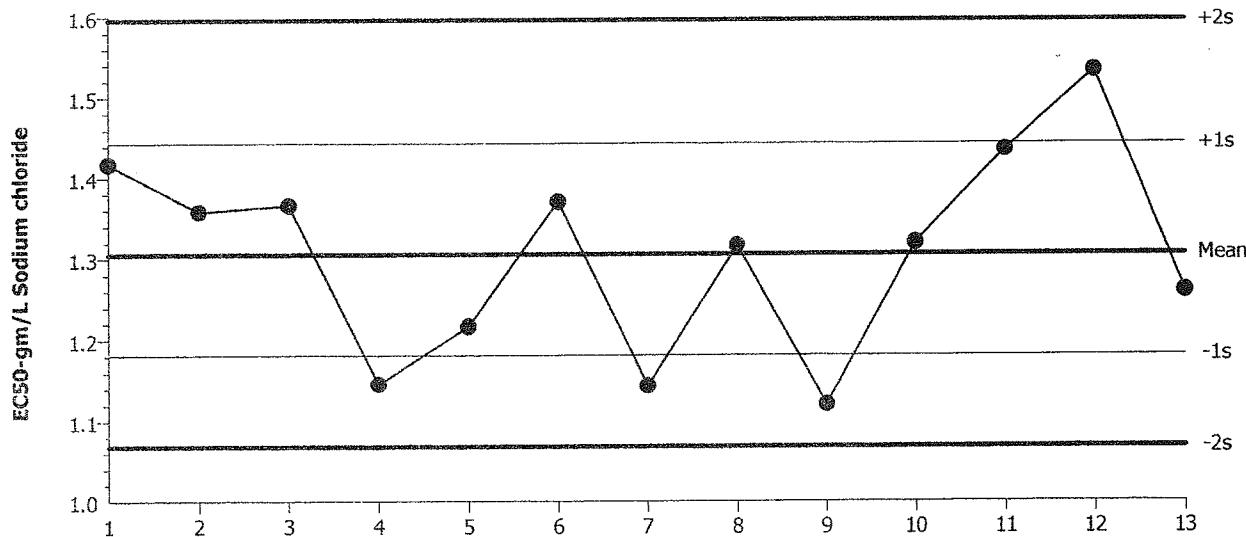
+2s Action Limit: 2.878

Quality Control Data

Point	Year	Month	Day	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2011	Nov	10	2.144	0.2157	0.5293			18-1646-5148	10-8016-4521
2			18	1.977	0.04919	0.1257			11-7820-4865	00-4433-4101
3			19	1.741	-0.1867	-0.5084			08-1813-0818	01-3437-6850
4			25	1.414	-0.5136	-1.546	(-)		12-3676-9409	20-8924-6879
5		Dec	2	1.32	-0.6083	-1.892	(-)		03-1806-9758	03-8081-7233
6	2012	Mar	20	1.741	-0.1867	-0.5084			04-4181-4802	06-8447-2630
7		Jun	26	2.297	0.3696	0.8751			18-3335-1100	10-1106-6325
8		Jul	17	2.144	0.2157	0.5293			02-9547-9197	16-5989-6607
9		Sep	18	2.267	0.3388	0.8078			20-3257-9401	11-6459-7205
10		Oct	30	1.741	-0.1867	-0.5084			14-7011-9138	01-0378-8759
11		Dec	11	2.462	0.5344	1.221	(+)		18-7111-4230	17-4716-3730
12	2013	Mar	12	2.297	0.3696	0.8751			13-9507-4728	11-5347-8239
13		Jun	18	1.866	-0.06177	-0.1625			19-2937-3586	14-8537-9739

Ceriodaphnia 7-d Survival and Reproduction Test			Rainier Environmental Laboratory		
Test Type:	Reproduction-Survival (7d)	Organism:	Ceriodaphnia dubia (Water Flea)	Material:	Sodium chloride
Protocol:	EPA/821/R-02-013 (2002)	Endpoint:	Reproduction	Source:	Reference Toxicant-REF

Ceriodaphnia 7-d Survival and Reproduction Test



Mean: 1.306 Count: 12 -1s Warning Limit: 1.181 -2s Action Limit: 1.069
Sigma: NA CV: 10.50% +1s Warning Limit: 1.444 +2s Action Limit: 1.596

Quality Control Data

Point	Year	Month	Day	QC Data	Delta	Sigma	Warning	Action	Test ID	Analysis ID
1	2011	Nov	10	1.417	0.1115	0.8175			18-1646-5148	02-8001-5569
2			18	1.358	0.05211	0.3905			11-7820-4865	14-3007-5816
3			19	1.366	0.05998	0.4482			08-1813-0818	00-3657-8188
4			25	1.146	-0.1597	-1.302	(-)		12-3676-9409	18-2334-7627
5		Dec	2	1.216	-0.08943	-0.708			03-1806-9758	11-1567-8111
6	2012	Mar	20	1.37	0.06434	0.4799			04-4181-4802	14-9819-7042
7		Jun	26	1.144	-0.162	-1.322	(-)		18-3335-1100	06-3063-9294
8		Jul	17	1.316	0.01035	0.07878			02-9547-9197	03-2978-8518
9		Sep	18	1.121	-0.1852	-1.527	(-)		20-3257-9401	21-2717-5233
10		Oct	30	1.319	0.01355	0.103			14-7011-9138	13-0927-4963
11		Dec	11	1.435	0.1293	0.9424			18-7111-4230	00-5310-2281
12	2013	Mar	12	1.533	0.2272	1.6	(+)		13-9507-4728	19-0889-8785
13		Jun	18	1.26	-0.04604	-0.3582			19-2937-3586	04-7875-5639

CETIS Summary Report

Report Date: 03 Jul-13 15:34 (p 1 of 2)

Test Code: RT061813CD | 19-2937-3586

Ceriodaphnia 7-d Survival and Reproduction Test				Rainier Environmental Laboratory	
Batch ID:	11-0456-7041	Test Type:	Reproduction-Survival (7d)	Analyst:	Eric Tollefson
Start Date:	18 Jun-13 16:00	Protocol:	EPA/821/R-02-013 (2002)	Diluent:	Mod-Hard Synthetic Water
Ending Date:	25 Jun-13 16:00	Species:	Ceriodaphnia dubia	Brine:	
Duration:	7d 0h	Source:	In-House Culture	Age:	<24h
Sample ID:	05-9836-4630	Code:	RT061813CD	Client:	Internal Lab
Sample Date:	18 Jun-13	Material:	Sodium chloride	Project:	
Receive Date:	18 Jun-13	Source:	Reference Toxicant		
Sample Age:	16h	Station:	In House		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
13-7392-8315	7d Survival Rate	2	4	2.828	NA		Fisher Exact/Bonferroni-Holm Test
09-8190-6493	Reproduction	0.5	1	0.7071	18.9%		Steel Many-One Rank Sum Test

Point Estimate Summary

Analysis ID	Endpoint	Level	gm/L	95% LCL	95% UCL	TU	Method
14-8537-9739	7d Survival Rate	LC50	1.866	1.445	2.41		Spearman-Kärber
04-7875-5639	Reproduction	IC5	0.5125	0.09207	0.5928		Linear Interpolation (ICPIN)
		IC10	0.5895	0.1926	0.6953		
		IC15	0.6704	0.4098	0.814		
		IC20	0.7554	0.5217	0.9337		
		IC25	0.8447	0.6595	1.029		
		IC40	1.102	0.8669	1.227		
		IC50	1.26	1.016	1.371		

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits	Overlap	Decision
13-7392-8315	7d Survival Rate	Control Resp	1	0.8 - NL	Yes	Passes Acceptability Criteria
14-8537-9739	7d Survival Rate	Control Resp	1	0.8 - NL	Yes	Passes Acceptability Criteria
04-7875-5639	Reproduction	Control Resp	34.7	15 - NL	Yes	Passes Acceptability Criteria
09-8190-6493	Reproduction	Control Resp	34.7	15 - NL	Yes	Passes Acceptability Criteria
09-8190-6493	Reproduction	PMSD	0.1894	0.13 - 0.47	Yes	Passes Acceptability Criteria

7d Survival Rate Summary

C-gm/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Dilution Water	10	1	1	1	1	1	0	0	0.0%	0.0%
0.25		10	1	1	1	1	1	0	0	0.0%	0.0%
0.5		10	1	1	1	1	1	0	0	0.0%	0.0%
1		10	0.9	0.7819	1	0	1	0.1	0.3162	35.14%	10.0%
2		10	0.5	0.3032	0.6968	0	1	0.1667	0.527	105.4%	50.0%
4		10	0	0	0	0	0	0	0		100.0%

Reproduction Summary

C-gm/L	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Dilution Water	10	34.7	32.25	37.15	17	40	2.071	6.55	18.88%	0.0%
0.25		10	35	32.35	37.65	16	41	2.246	7.102	20.29%	-0.86%
0.5		10	33.4	31.14	35.66	17	38	1.916	6.059	18.14%	3.75%
1		10	23.3	20.16	26.44	0	28	2.659	8.407	36.08%	32.85%
2		10	3.8	2.218	5.382	0	9	1.34	4.237	111.5%	89.05%
4		10	0	0	0	0	0	0	0		100.0%

CETIS Summary Report

Report Date: 03 Jul-13 15:34 (p 2 of 2)

Test Code: RT061813CD | 19-2937-3586

Ceriodaphnia 7-d Survival and Reproduction Test**Rainier Environmental Laboratory****7d Survival Rate Detail**

C-gm/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Dilution Water	1	1	1	1	1	1	1	1	1	1
0.25		1	1	1	1	1	1	1	1	1	1
0.5		1	1	1	1	1	1	1	1	1	1
1		1	1	1	1	1	1	1	1	1	0
2		1	1	0	1	0	0	0	1	0	1
4		0	0	0	0	0	0	0	0	0	0

Reproduction Detail

C-gm/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Dilution Water	37	40	39	37	35	35	33	38	36	17
0.25		36	38	36	41	37	37	37	40	32	16
0.5		35	38	38	34	37	35	34	33	33	17
1		25	22	28	25	25	28	27	28	25	0
2		4	8	0	9	0	0	0	9	0	8
4		0	0	0	0	0	0	0	0	0	0

7d Survival Rate Binomials

C-gm/L	Control Type	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	Dilution Water	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
0.25		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
0.5		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
1		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	0/1
2		1/1	1/1	0/1	1/1	0/1	0/1	0/1	1/1	0/1	1/1
4		0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1

Rainier Environmental
Washington Laboratory

Client: Reference Test
Sample ID: 4.0g/L NaCl
Test No: RT061913CD
Log-In#:

Initial and Final Chemistries

Seven Day Chronic Freshwater Bioassay

Start Date & Time: 6/18/13 1600

Stop Date & Time: 6/25/13 1600

Test Species: Ceriodaphnia dubia

Conc. or % <u>CON</u>	Days													
	0		1		2		3		4		5		6	
	init.	final												
pH	7.65	7.82	7.57	7.61	7.63	7.72	7.61	7.57	7.45	7.61	7.62	7.81	7.58	7.65
DO (mg/l)	8.1	8.2	7.9	8.1	7.7	8.1	7.9	8.2	7.9	8.2	7.7	8.1	7.9	8.2
Cond. (μmhos-cm)	190	192	195	191	187	199	191	203	189	207	186	205	188	195
Temperature (°C)	24.2	25.1	24.7	25.5	24.3	25.7	24.8	25.4	25.0	25.2	24.8	25.4	24.9	25.2
<u>0.25</u>	Days													
	0		1		2		3		4		5		6	
	init.	final												
pH	7.68	7.91	7.58	7.64	7.55	7.75	7.63	7.55	7.48	7.72	7.63	7.77	7.55	7.73
DO (mg/l)	8.1	8.2	8.0	8.2	7.7	8.1	8.0	8.2	7.9	8.2	7.6	8.0	8.1	8.2
Cond. (μmhos-cm)	657	663	645	657	662	659	653	661	655	663	647	659	661	673
Temperature (°C)	24.2	25.1	24.7	25.5	24.4	25.5	24.5	25.4	25.0	25.5	24.8	25.4	24.9	25.4
<u>0.5</u>	Days													
	0		1		2		3		4		5		6	
	init.	final												
pH	7.58	7.87	7.59	7.66	7.53	7.77	7.57	7.63	7.52	7.73	7.65	7.79	7.57	7.73
DO (mg/l)	8.2	8.2	8.0	8.2	7.8	8.1	8.0	8.2	7.8	8.2	7.6	8.0	8.0	8.2
Cond. (μmhos-cm)	1207	1215	1198	1215	1205	1245	1213	1231	1215	1228	1205	1235	1221	1247
Temperature (°C)	24.3	25.1	24.8	25.6	24.2	25.4	24.5	25.4	25.0	25.5	24.7	25.3	24.8	25.4
<u>1.0</u>	Days													
	0		1		2		3		4		5		6	
	init.	final												
pH	7.62	7.90	7.57	7.62	7.54	7.78	7.58	7.65	7.51	7.75	7.66	7.79	7.55	7.75
DO (mg/l)	8.1	8.2	7.6	8.2	7.8	8.2	8.1	8.1	7.9	8.2	7.5	8.0	8.1	8.2
Cond. (μmhos-cm)	2123	2135	2096	2113	2135	2145	2117	2137	2122	2142	2124	2151	2151	2185
Temperature (°C)	24.3	25.1	24.8	25.5	24.2	25.3	24.8	25.4	25.0	25.4	24.7	25.3	24.7	25.5
<u>2.0</u>	Days													
	0		1		2		3		4		5		6	
	init.	final												
pH	7.61	7.92	7.57	7.58	7.55	7.81	7.58	7.68	7.51	7.75	7.67	7.80	7.55	7.76
DO (mg/l)	8.0	8.1	7.9	8.2	7.7	8.2	7.8	8.2	7.8	8.2	7.5	8.1	8.0	8.2
Cond. (μmhos-cm)	3990	4030	3890	3970	3950	4070	4010	4070	3970	4030	3980	4050	4030	4100
Temperature (°C)	24.4	25.1	24.8	25.5	24.3	25.3	24.8	25.4	25.0	25.4	24.7	25.3	24.8	25.5
<u>4.0</u>	Days													
	0		1		2		3		4		5		6	
	init.	final												
pH	7.60	7.91												
DO (mg/l)	8.0	8.2												
Cond. (μmhos-cm)	7670	7710												
Temperature (°C)	24.4	25.1												
Tech. Initials	BT													

Dilution Water Batch #: DMW 004
Test Chamber: VWR

QA Check: _____

Sample Description: 4.0g/L NaCl
Animal Source: In House Culture
Comments: _____

Date Received: _____ Date of Hatch: _____

Ceriodaphnia 7-Day Chronic Survival and Reproduction

Start Date and Time: 6/18/13 1600
Stop Date and Time: 6/25/13 1600

Client/Sample ID: Reference Testout RTD61812CD
Test Number:

Rep	Daily Reproduction								Day 6 Total	Third Brood
	1	2	3	4	5	6	7	8		
1	CON	19	—	—	5	—	12	20	17	37
2		27	—	—	6	—	13	21	19	40
3		29	—	—	1	4	—	15	20	19
4		57	—	—	—	5	—	12	20	17
5		3	—	—	—	5	11	17	19	39
6		48	—	—	—	5	—	10	20	15
7		49	—	—	—	5	—	10	18	15
8		54	—	—	—	6	—	12	20	15
9		15	—	—	—	5	—	11	19	17
10		10	—	—	—	5	—	12	—	17
Analyst	gt	94	gt	gt	gt	gt	gt	gt	gt	gt
Time	1600	115	1450	1100	115	115	130	1600		
Selen #	003	003	003	003	003	003	003	003		
Rep	Cone.	Cont	1	2	3	4	5	6	Day 6 Total	Third Brood
1	0.35	30	—	—	6	—	12	18	16	35
2		44	—	—	5	—	12	21	17	39
3		37	—	—	5	—	11	20	16	36
4		16	—	—	6	—	13	22	19	41
5		7	—	—	6	—	12	19	13	37
6		50	—	—	5	—	12	20	17	37
7		46	—	—	5	—	13	21	17	37
8		59	—	—	4	—	10	18	14	32
9		8	—	—	—	5	—	11	16	16
10										

Rep	Daily Reproduction								Day 6 Total	Third Brood
	1	2	3	4	5	6	7	8		
1	CON	19	—	—	5	—	12	20	17	37
2		27	—	—	6	—	13	21	19	40
3		29	—	—	1	4	—	15	20	19
4		57	—	—	—	5	—	12	20	17
5		3	—	—	—	5	11	17	19	39
6		48	—	—	—	5	—	10	20	15
7		49	—	—	—	5	—	10	18	15
8		54	—	—	—	6	—	12	20	15
9		15	—	—	—	5	—	11	19	17
10		10	—	—	—	5	—	12	—	17

Rep	Daily Reproduction								Day 6 Total	Third Brood
	1	2	3	4	5	6	7	8		
1	0.5	38	—	—	4	—	11	20	15	35
2		32	—	—	6	—	13	19	19	38
3		53	—	—	6	—	12	20	15	38
4		55	—	—	5	—	11	18	16	34
5		58	—	—	5	—	11	20	16	37
6		12	—	—	5	—	12	18	17	35
7		17	—	—	4	—	10	20	14	34
8		19	—	—	4	—	9	20	13	33
9		2	—	—	4	—	10	19	14	33
10		43	—	—	5	—	12	—	17	17

Comments:

X=mortality

Appendix D
Chain-of-Custody Forms



Chain of Custody

Washington
5013 Pacific Highway East, Suite 20 Flite,
WA 98424
Phone 253.922.8898

Date _____ Page _____ of _____

Sample Collection By: Dawnick Perry

Report to:

Company

Address

City/State/Zip

Contact

Phone

Email

Washington Beef LLC
201 Edmund Rd
Tacoma, WA 98448
Shelly Byers, Eddy
Ses 1352, 6534

Invoice To:

Company

Address

City/State/Zip

Contact

Phone

Email

Receipt Temperature (°C)

WET

ANALYSES REQUIRED

SAMPLE ID	DATE	TIME	MATRIX	CONTAINER TYPE	NO. OF CONTAINERS	COMMENTS	10
Final Effluent	6/17/13	2153m 6th	Cubicleter	1			
2							
3							
4							
5							
6							
7							
8							
9							
10							

PROJECT INFORMATION

SAMPLE RECEIPT

RELINQUISHED BY (CLIENT)

RELINQUISHED BY (COURIER)

Client:	Total No. of Containers	1	(Signature)	(Time)	(Signature)	(Time)	(Signature)	(Time)
PO No.:	Received Good Condition?	✓	(Printed Name)	(Date)	(Printed Name)	(Date)	(Printed Name)	(Date)
Shipped Via:	Matches Test Schedule?	✓	(Company)	(Company)	(Company)	(Company)	(Company)	(Company)
SPECIAL INSTRUCTIONS/COMMENTS:								

RECEIVED BY (COURIER)

RECEIVED BY (LABORATORY)

(Signature)	(Time)	(Signature)	(Time)
(Printed Name)	(Date)	(Printed Name)	(Date)
(Company)	(Log In #)	(Company)	(Log In #)

13-061

Eric Tolleson 1415
(Printed Name) (Date)
ERIC TOLLESON 6/18/13
(Company)

Rainier ENVIRONMENTAL

Washington
5013 Pacific Highway East, Suite 20 Fife,
WA 98424
Phone 253.922.8898

Date 01/19/13 Page 1 of 1

Chain of Custody

Sample Collection By:

Report to:
Company Washington Beef LLC
Address 501 Elmwood Rd
City/State/Zip Tottenish, WA 98948
Contact Sherri Beers
Phone 509-952-6534
Email Sherry.Beers@abbeef.com

Invoice To:

Company Same
Address _____
City/State/Zip _____
Contact _____
Phone _____
Email _____

ANALYSES REQUIRED

SAMPLE ID	DATE	TIME	MATRIX	CONTAINER	NO. OF CONTAINERS	COMMENTS
1	EF001913	7:15 AM	West-Wake Crust	1	1.0	
2						
3						
4						
5						
6						
7						
8						
9						
10						

ET
J

Receipt Temperature (°C)

PROJECT INFORMATION	SAMPLE RECEIPT	RELINQUISHED BY (CLIENT)	RELINQUISHED BY (COURIER)
Client:	Total No. of Containers	(Signature) <u>Sherry Beers</u> (Printed Name) <u>12:00pm</u> (Date) <u>1/19/13</u> (Company)	(Signature) (Printed Name) (Date) (Company)
PO No.:	Received Good Condition?	Y	
Shipped Via:	URS	Matches Test Schedule?	Y

RECEIVED BY (COURIER)	RECEIVED BY (LABORATORY)
(Signature) (Printed Name)	(Signature) (Printed Name)
(Time)	(Time)
Eric Tolosa 6/20/13	1400 (Log in #)

SPECIAL INSTRUCTIONS/COMMENTS:

(Signature)	(Signature)
(Printed Name)	(Printed Name)
(Date)	(Date)
(Company)	(Company)

13-054



Washington
5013 Pacific Highway East, Suite 20 Fife,
WA 98424
Phone 253.922.8898

Chain of Custody